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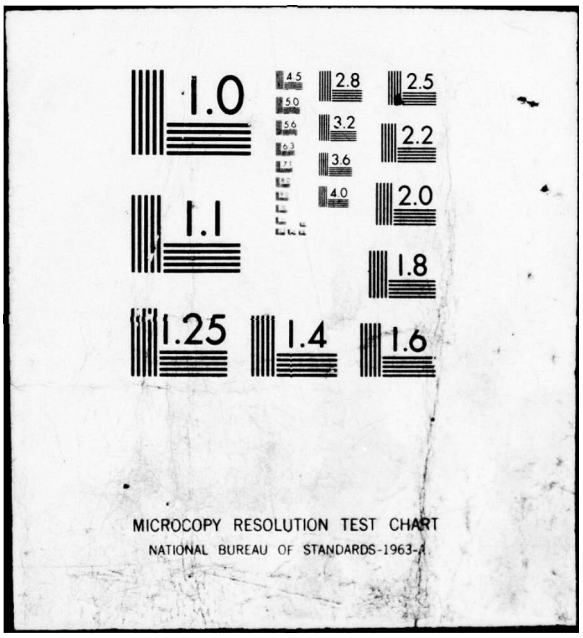
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GENETIC CONTROL OF THE GERMAN COCKROACH

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The overall goal of this Contract was to investigate the possibility of controlling the German cockroach, Blattella germanica (L.), by the use of chromosome translocations. Translocation stocks and mutant markers requisite to the proposed research were already available.

The first objective

Laboratory developmental studies were conducted according to three specific objectives. The first involved the study of wild-type laboratory

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and field populations. The origin, hatch, and development of nymphal groups that determine the growth and age structure of a laboratory population were analyzed. In field collections, nymphs formed an unexpectedly small segment of the populations. Their distribution/instar was relatively even. Inhibition of reproduction through limited supplies of food and water could account for this pattern. The data suggested that the effectiveness and size of the releases might vary with the season and/or particular environmental conditions. Properties of 9 single translocations were studied under Objective 2. Eight showed good competitiveness, 1 marginal, 1 poor, and 2 were not tested due to insufficient time. Eight were established in backcross systems to closely linked markers for maintenance and identification. Population studies using repeated releases of translocation-carrying males were conducted under Objective 3. Single translocations retarded but did not suppress population growth. An unusual interchange involving 3 chromosomes approached suppression, but it required a double translocation to bring about a decline in total numbers. Thus, the scope of Objective 3 was widened to include the synthesis and study of double translocations. Fully competitive double males were developed, with sterilities ranging from 70-90%. Time limits prohibited population studies with the males characterized by 90% sterility. A 4th objective, field trials, was mentioned in the grant proposal. These were to be undertaken if it was possible to complete the laboratory developmental studies. The time allowed was not sufficient to undertake this objective.

The authors found that

The sterility effect, "embryonic trapping", that is used in population suppression by double translocations is due to a reduction in the numbers of viable embryos/ootheca. The frequency of sterile egg cases increases sharply at lethalities of 60-80%, with complete sterility at lethalities of 80% or better. We conclude that double translocations are the most effective method of using translocations for control in B. germanica. Such mechanisms are potentially capable of reducing field populations.

A

Genetic Control of the German Cockroach
(Final report on contract N00025-74-e-1014, 3/1/74-2/28/77)

Mary H. Ross and Donald G. Cochran

The purpose of this Contract was to investigate the potential usefulness of reciprocal chromosome translocations for control of the German cockroach, Blattella germanica (L.). Basic genetic and cytogenetic research had been underway in our laboratories for many years. As a result, we had isolated a number of translocation-carrying lines, as well as mutant markers which were expected to provide a means of identifying the translocation-carrying individuals. It also appeared the species had attributes which would make it a favorable candidate for control by genetic means.

The research accomplished under this Contract is summarized according to each of the objectives stated in the original proposal. This is followed by a brief discussion of the more important findings and their implications for prospective control efforts.

Objective 1 - Population structure of wild-type "field" and laboratory populations.

Gross estimates of the size of cockraoch infestations are available from published reports. The effects of overcrowding on laboratory populations of the American cockroach have also been described. However there is a dearth of information concerning the age distribution within either free or laboratory populations, not to mention factors that control the growth and structure of free populations. A detailed analysis of the growth of laboratory populations, as well as a preliminary study of some field collections, was completed on the German cockroach under this Contract.

A. Field collections - Collections from 9 low income homes in Raleigh, N.C., were made available to us through the cooperation of Dr. Charles G. Wright of North Carolina State University. The collections were made in the summer of 1973 by use of a modified suction vacuum cleaner. Collection was continued until repeated search gave negative results. It was felt they represented a reasonably complete sample of the resident populations.

The results of analyzing the collections for age distribution and, in late instars, for sex, are shown in Table 1. These have several implications of significance for field trials, as noted below:

(1) Numbers of late instar nymphs were low. Releases of less than 500 nymphs of matching age would reach ratios of 10T:1 normal male. This should suppress the productivity of future adult groups, but not that of the rather large segment of the populations already in the adult stage. Continued low level hatch from the latter group would depend on whether density-controlling factors remained as restrictive as they obviously were earlier (see below).

(2) Oothecal examination indicated populations were on the verge of a considerable increase in hatch. This agreed with field observations of Wright who has reported that populations peak in late-summer to early fall.

To be most effective, it seems releases should be timed so that the sterility factor was incorporated into this breeding group, i.e., possibly by starting releases in late winter-early spring. Alternatively, an integrated approach using insecticides for reduction of adult numbers might be used.

(3) Productivity, measured from ratios of adult females/nymphs (Table 1, col. 11), was low in comparison to that seen in the laboratory populations. The sole exception was Coll. 9U, which seemed to represent a specialized case (see #5). Populations growth had clearly been severely restricted. The effects of releases into such highly regulated populations would depend, in part, on the nature of the regulating factors (see #4).

(4) The age analyses showed a marked tendency toward a uniform distribution of nymphs throughout the 6 instars (except see #5). There was no consistent trend toward a reduction in number with age. Thus, it appeared that the low numbers of nymphs might be due to a lack of reproduction, rather than nymphal mortality. This theory is supported by certain physiological characteristics of the cockroach. It is known that starved females will not mate, that preovipositional feeding is a requirement for oothecal production, and that water intake must be sufficient for both the body and developing eggs. Inhibition of oviposition would occur before genetic death or sterility from embryonic trapping, and hence the effects would be additive. This would be an asset to genetic control measures in populations regulated by such factors.

(5) The large numbers of small nymphs in Coll. 9U may be evidence of the importance of water availability to hatch. This was a utility room containing buckets of water and damp mops. It is probable females migrated to this area just prior to hatch - a behavioral trait we have frequently observed in the laboratory.

Natural populations can be expected to vary with locale, immediate environmental conditions, probably with season, and of course, as a result of control measures and re-introductions. We still know very little about density-dependent regulation in cockroach populations. The analyses of Wright's collections highlighted our need for a year-round study of population dynamics and the importance of analyzing samples from proposed release sites prior to field testing. The most rapid suppression would be obtained where, through seasonal variations, chemical applications, or a combination of both, there were the fewest numbers of adult females within the resident population. Since adult females generally mate once within their lifetime, it is obviously desirable that mating involves a sterile male - a goal that cannot be reached for previously mated adults.

B. Laboratory populations. Growth of laboratory wild-type populations was studied using two experimental designs. In the first, a freely intrabreeding population was censused at monthly intervals for a period of 7 months (Ross 1975). The results are shown in Fig. 1 (Control). Population growth was analyzed insofar as discrete progeny groups could be distinguished. Hatch and development of these groups is shown diagrammatically in Fig. 2. As can be seen, there was overlapping hatch of certain groups, such as the F_2(1-2) and F_2(2-1), in later months. The designation system was devised in order to refer to nymphal groups from successive egg cases of the same parental group and also those from different parental groups. Thus, F_1-1, F_1-2, F_1-3, etc. = nymphs from

Table 1. Analyses of the age structure of "field" collections of B. germanica.

House	Room[a]	Total no.	No. adults (♂,♀)	No. per instar 6 (♂,♀)	5 (♂,♀)	4	3	2	1	Ratio adult ♀: nymphs	Notes on sanitation
1	K	23	3 (2,1)	4 (1,3)	4 (2,1)	0	0	0	0	1:8	good
2	K	196	114 (35,79)	32 (13,19)	31 (11,20)	28	27	42	42	1:3	poor
3	K	223	141 (72,69)	28 (18,10)	4 (3,1)	25	9	2	14	1:2	good
4	K	183	74 (32,42)	11 (4,7)	17 (8,9)	27	17	11	26	1:3	avg
4	L	49	11 (4,7)	11 (6,5)	1 (0,1)	7	3	5	11	1:5	
4	B	16	2 (0,2)	1 (1,0)	0	0	1	1	11	1:7	
5	K	426	220 (100,120)	34 (12,22)	26 (7,19)	39	39	25	53	1:2	avg
6	K	521	102 (53,49)	81 (31,50)	61 (33,28)	131	79	49	18	1:8	poor
7	K	685	193 (79,114)	64 (23,41)	39 (17,22)	101	78	126	84	1:4	poor
7	L	167	62 (36,26)	11 (8,3)	19 (10,9)	16	22	15	22	1:4	
7	B	8[illegible]	53 (23,30)	21 (11,10)	4 (2,2)	5	2	0	0	1:1	
8	K	939	347 (194,153)	11 (10,1)	121 (50,71)	74	63	112	211	1:4	very poor
8	B (1st)	79	59 (41,18)	9 (3,6)	1 (1,0)	1	0	6	3	1:1	
8	B (2nd)	250	67 (30,37)	50 (26,24)	14 (6,8)	30	12	19	58	1:5	
9	K	644	356 (182,174)	96 (49,47)	55 (26,29)	52	33	24	27	1:2	very poor
9	U	1445	114 (13,101)	24 (6,18)	36 (13,23)	93	244	385	549	1:13	

[a] Letters indicate sites, i.e., kitchen (K), living room (L), bedroom (B), and utility room (U).

1st, 2nd, 3rd and later egg cases of the parental (P_1) group; F_2(1-1), F_2 (1-2), F_2 (1-3)..= nymphs from sucessive egg cases of the first F_1 group, i.e., the F_1-1); F_2 (2-1), F_2 (2-2), F_2 (2-3)..= nymphs from successive egg cases of the 2nd F_1 group, i.e., the F_1-2. The system is repeated for other nymphal groups.

The data showed that periods of rapid growth marked the initial appearance of a new generation, in contrast to a slower expansion occasioned by hatch of successive oothecae from groups belonging to the same generation. Nymphal groups (F_2) from 1st to 3rd egg cases were approximately 12X, 11X, and 8X larger than their parental groups. Those from later oothecae made negligible contributions to population growth. The ever-increasing numbers of groups hatching within the population and their staggered pattern explains both the manner in which a population grows and why older populations have nymphs in all age groups.

The second type of population experiment used isolated progeny groups. It was felt this method might improve on the use of a freely intrabreeding population in that (1) mortality among small nymphs that occurred from overcrowding in late moths in the 1st experiment could be avoided and (2) progeny groups could be measured that were indistinguishable due to overlapping hatch, again in the later months of the first experiment.

The method consisted of separating each nymphal group from its parents, and keeping records as to hatch, development and egg case production (Ross 1976). These data made it possible to reconstruct the total population for each month of growth (Table 2). This was probably as good a measure of the reproductive potential of the species as one could obtain with a population-type experiment. Counts of the individual nymphal groups are shown in Table 3, +/+. Comparison between groups belonging to different generations (F_1, F_2 and F_3) shows why growth was accelerated with the initial hatch of a new generation (Tables 2 and 4 in mo. 4 and 7). In the group situation, females averaged 31.3 $\pm$ 0.5 nymphs from 1st and 2nd ootheca, in contrast to 40.0 $\pm$ 1.0 in single-pair matings. This dropped to 29.8 $\pm$ 0.5 in 3rd oothecae. Only 3 groups from 4th or 5th ootheca, in hatched within the 7 mo. of study, and within these female mortality was highly variable. No meaningful estimate of hatch was obtained other than a rough estimate of ca. 7 nymphs from 5th oothecae. Nymphal survival was estimated at 83 percent, adult males generally died within 90-120 days; females seldom survived over 5 months. Other biological parameters useful in analyzing and/or predicting population growth are summarized in Table 5.

The laboratory population experiments provided the control data whereby the effects of releasing single and double translocation-carrying males were determined (Objective 3). They illustrate how a population might undergo very rapid growth if, for example, it was reduced to below the carrying-capacity of its environment by insecticide application (non-residual). The data will also be useful in planning the temporal sequence of releases. From these studies and the field collections, it seems highly probable we will find nymphs of all age groups within most free populations.

Fig. 1. Growth of an experimental and a wild type population of B. germanica as determined from monthly censuses. Age is indicated by "Ad" for adult and "I 1-6" for the six nymphal instars.

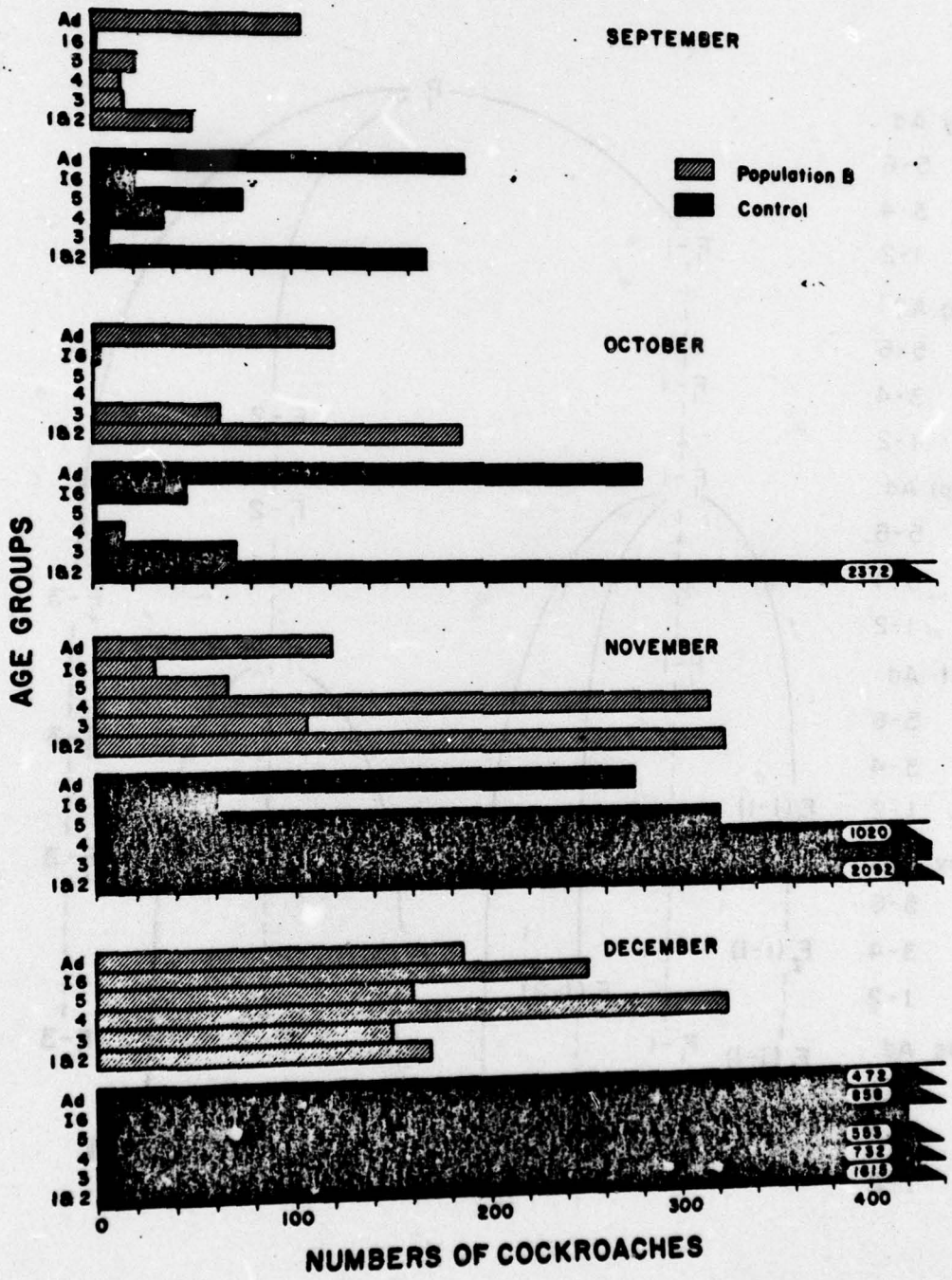

Fig. 2. The origin and growth of progeny groups seen in population studies of B. germanica. Solid lines connect parental with progeny groups produced from successive egg cases. Dotted lines show the approximate growth/month of each nymphal group. Ages are divided as to Ad (adults), instars 5 and 6 (large nymphs), instars 3 and 4 (medium-sized nymphs), and instars 1 and 2 (small nymphs).

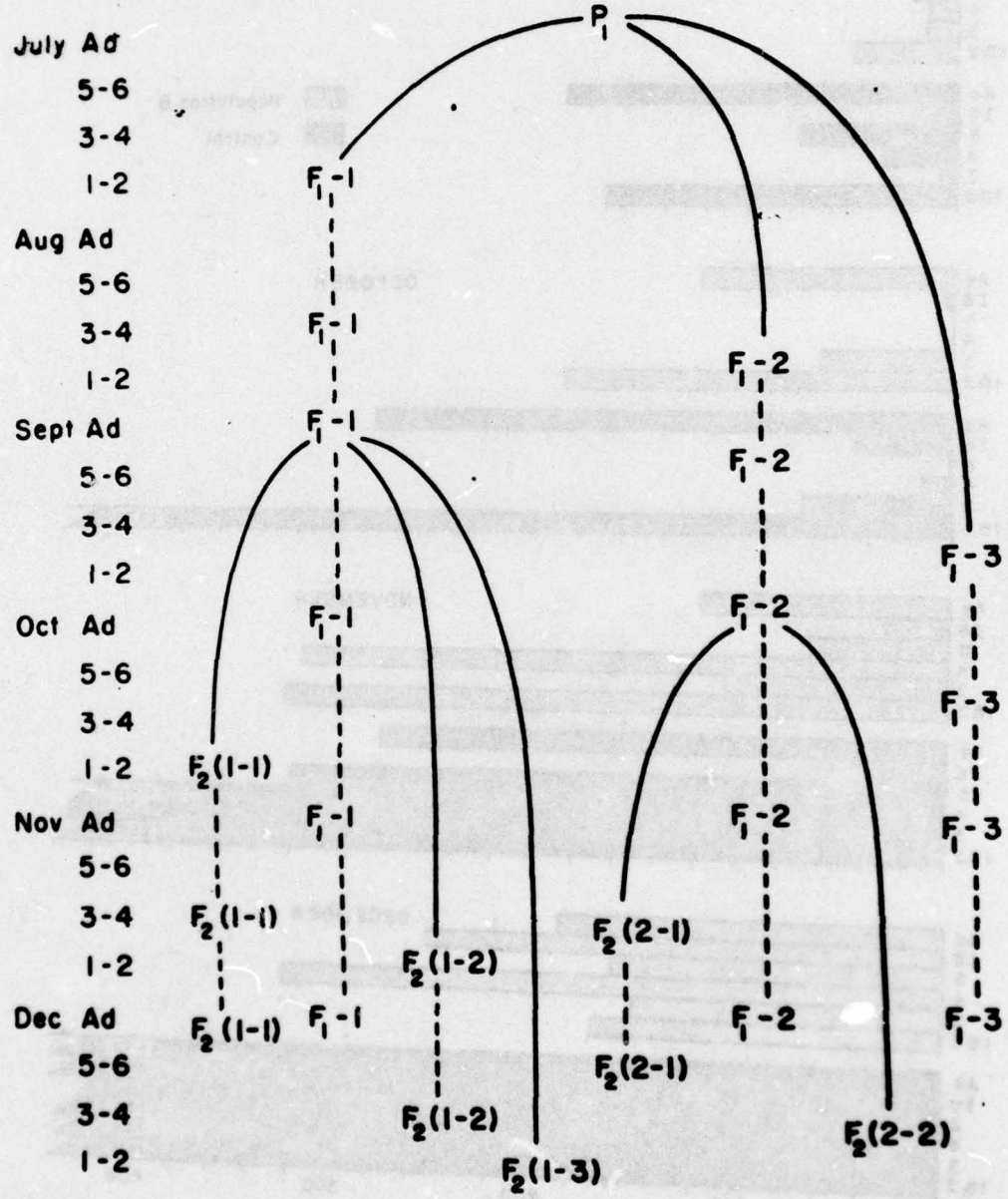

Table 2. Population[a] growth and structure from experiments with wild-type (+/+) B. germanica.

Age[b]	Month of growth						
	1	2	3	4	5	6	7
Ad	10	10	178	342	463	2568	6573
Lg		168	168	131	2105	4105	5861
Md							
Sm	203	202	158	2536	4946	7062	38957[c]
Total	213	380	504	3009	7514	13735	51391

[a]Population reconstructed from isolated progeny groups. See text for explanation.

[b]Age is indicated by "Ad" for adult and "Lg" for large (instars 5-6) "Md" for medium (instars 3-4) and "Sm" for small (instars 1-2) nymphs. Nos. of adults are estimated as 2X the no. of adult females.

[c]Includes 1 group, the F_3 (1-1-1), which was estimated on the basis of hatch data.

Table 3 - Progeny groups from experiments with wild type, T(?;9), +/st and T(4;8;10) German cockroaches.

Progeny group	+/+		T(8;9),+/st			T(4;8;10)			
	A	B	A	B	C	A	B	C	D
F_1-1	209	198	78	107	80	34	11	43	47
F_1-2	195	208	83	112	34	15	45	62	31
F_1-3	110	193	84	99	59	27	14	41	14
F_1-4	59	43	33	34	19	11	13	16	0
F_1-5	26	14	13	9	4	2	0	3	0
F_2(1-1)	2490	2582	512 (53 st)	656 (62 st)	118 (18 st)	101	42	152	127
F_2(1-2)	2535	2862	536 (41 st)	607 (58 st)	154 (21 st)	118	24	86	68
F_2(1-3)	2287	2374	398 (28 st)	498 (39 st)	82 (11 st)	79	23	42	50
F_2(1-4)	1415	1855	231 (16 st)	362 (28 st)	17 (2 st)	61	17	65	27
F_2(1-5)[a]	544	655							
F_2(2-1)	2444	2352	485 (36 st)	581 (20 st)	57 (2 st)	117	119	114	58
F_2(2-2)	2682	2202	446 (42 st)	550 (35 st)	51 (3 st)	123	115	94	48
F_2(2-3)[a]	2332	2074	412 (33 st)	439 (19 st)	23 (1 st)	134	37	106	19
F_2(3-1)	1693	2888	674 (70 st)	483 (31 st)	222 (27 st)	71	52	73	28
F_2(3-2)	1453	2886	640 (51 st)	423 (13 st)	157 (24 st)	127	54	61	14
F_2(3-3)[a]	1206	1996	523 (43 st)	321 (15 st)	96 (15 st)	87	12	16	--
F_2(4-1)	871	523	191 (11 st)	166 (12 st)	37 (2 st)	22	41	38	0
F_3(1-1-1)	33370[b]	33569[b]	2198 (153 st)	2611 (124 st)	451 (96 st)	595	65	386	135

[a] Groups that hatched later than the 7th mo.

[b] Estimated as 13X the appropriate F_2(1-1) group.

Table 4. - Growth rates of wild-type (+/+) and experimental populations[a] of B. germanica.

Experiment	Group	Month 2	3	4	5	6	7
+/+	sm nymphs	1.0X	<1.0X	16.0X	2.0X	1.4X	5.5X
	total	1.8X	1.3X	6.0X	2.5X	1.8X	3.7X
T(?;9),+/st: avg. A-B	sm nymphs	1.0X	<1.0X	6.3X	1.8X	1.4X	2.4X
	total	1.8X	1.4X	3.2X	2.1X	1.7X	2.2X
C	sm nymphs	<1.0X	1.7X	2.0X	1.8X	1.7X	1.8X
	total	1.2X	1.5X	1.7X	1.7X	1.7X	1.7X
T(4;8;10): avg. A-C	sm nymphs	1.4X	<1.0X	4.3X	1.7X	1.1X	2.6X
	total	1.9X	1.3X	2.2X	1.8X	1.5X	1.9X
D	sm nymphs	<1.0X	<1.0X	9.1X	<1.0X	<1.0X	1.5X
	total	1.4X	1.1X	2.5X	1.5X	1.3X	1.6X

[a] Reconstructed from isolated progeny groups. See text for explanation.

Table 5. Observations of nymphal development and hatch from isolated progeny groups of German cockroaches.

Period measured	Means ± s.e. and range (days)
Nymphal development (hatch to maturation)	53.9 ± 2.1 (42-65)
Maturation to hatch of 1st oothecae	33.4 ± 1.1 (30-38)
Hatch of 1st to hatch of 2nd oothecae	32.8 ± 1.3 (29-38)
Hatch of 2nd to hatch of 3rd oothecae	33.3 ± 1.1 (30-38)
Hatch of the F_1 to hatch of the F_2	89.3 ± 2.8 (81-97)

Objective 2 - Studies of single translocation heterozygotes

At the time this Contract was initiated, we had made detailed studies of 3 out of a total of 20 reciprocal translocations, namely T(2;11)Cu, T(9;10)Pw, and T(8;9)2c (=T(9;11)2c of early reports (Table 6). It was of paramount importance to analyze additional stocks, for only then could we select those of most promise for suppression experiments and/or the development of double translocation stocks (see objective 3).

The primary requisite for usefulness was to find a closely linked mutant marker which could be used for identifying the translocation-carrying cockroaches. Chromosome measurements at late pachytene were used as an indication of the chromosomes involved in the translocation. Since linkage group-chromosome correlations were at least partially completed, it was often possible to reduce the number of tests by using markers believed to lie on the chromosomes involved in that particular translocation. Closely linked markers were found for 9 stocks, 7 of which are listed in Table 6, col. 3 (all except the first 3). Two others, T(2;11)2a and T(2;11)2i, showed close linkage with ru, but other characteristics of these stocks have not been studied. One stock, T(4;5?)1d, did not show linkage with any of the markers tested. All marked stocks are now in backcross systems to the most closely linked mutant. In all except T(11;12), at least 95% of the phenotypically normal progeny in each generation are translocation heterozygotes (T/+). T(11;12) is maintained by backcrosses to fs/fs females in which 91% are T/+.

Translocation identification made it possible to study other characteristics. Possibly the most important for genetic control prospects is male competitiveness. This was mainly tested by caging on T/+ and 1 normal male, aged 3-6 days post-maturation, with one normal female aged 4-6 days postmaturation (aged for maximum receptivity of female and sexual maturity of male). In most, the T/+ males competed equally or possibly somewhat better than the wild-type males (Table 6, col. 2). Only two, T(6;8)7h and T(2;11)Cu were markedly less competitive.

In translocation heterozygotes, random chromosome disjunction is expected to result in a 1:1 ratio of alternate (balanced chromosome complement) to adjacent (lethal = unbalanced) disjunction. This is reflected in the percent hatch. As shown in Table 6, cols. 4 and 5, hatch estimates of ca. 50% characterized translocations with random disjunction. Certain stocks showed a favoring of the alternate type of disjunction and hatch was raised accordingly. The close correlation between disjunction and hatch in males of most stocks makes it very likely the disjunction situation in females is also indicated by the hatch data. Good meiobic cells have not been found in females. Two stocks, T(8;9)2c and T(7;12), apparently show sexual dimorphism in disjunction but in opposite directions.

Certain aspects of the cytogenetic data have implications for basic research which have not yet been fully explored. For example, there may be a relationship between chromosome breakpoint position and the production of unusual sex differences in recombination (Ross and Cochran 1975). These may, in turn, be related to sex differences in disjunction. The analysis of certain cases of altered disjunction frequencies may lead to a fuller

Table 4. - Growth rates of wild-type (+/+) and experimental populations[a] of B. germanica.

Experiment	Group	Month 2	3	4	5	6	7
+/+	sm nymphs	1.0X	<1.0X	16.0X	2.0X	1.4X	5.5X
	total	1.8X	1.3X	6.0X	2.5X	1.8X	3.7X
T(?;9),+/st: avg. A-B	sm nymphs	1.0X	<1.0X	6.3X	1.8X	1.4X	2.4X
	total	1.8X	1.4X	3.2X	2.1X	1.7X	2.2X
C	sm nymphs	<1.0X	1.7X	2.0X	1.8X	1.7X	1.8X
	total	1.2X	1.5X	1.7X	1.7X	1.7X	1.7X
T(4;8;10): avg. A-C	sm nymphs	1.4X	<1.0X	4.3X	1.7X	1.1X	2.6X
	total	1.9X	1.3X	2.2X	1.8X	1.5X	1.9X
D	sm nymphs	<1.0X	<1.0X	9.1X	<1.0X	<1.0X	1.5X
	total	1.4X	1.1X	2.5X	1.5X	1.3X	1.6X

[a] Reconstructed from isolated progeny groups. See text for explanation.

Table 5. Observations of nymphal development and hatch from isolated progeny groups of German cockroaches.

Period measured	Means ± s.e. and range (days)
Nymphal development (hatch to maturation)	53.9 ± 2.1 (42-65)
Maturation to hatch of 1st oothecae	33.4 ± 1.1 (30-38)
Hatch of 1st to hatch of 2nd oothecae	32.8 ± 1.3 (29-38)
Hatch of 2nd to hatch of 3rd oothecae	33.3 ± 1.1 (30-38)
Hatch of the F_1 to hatch of the F_2	89.3 ± 2.8 (81-97)

Table 6. Characteristics of single translocation heterozygotes.

Stock	♂ competitiveness	Linkage estimates[a] and, if known, the chromosome carrying the mutant marker	Hatch (%)	Disjunction[b]	Homozygote
T(2;11)Cu	poor	----	♂ 49.5	♂ random	no test - Cu ♀ sterile
T(9;10)Pw	good[c]	r(Ch 10):2.6 ± 0.7% ro(Ch 10):0.8 ± 0.1% ru(Ch 9):complete st(Ch 9):3.5 ± 0.8% (♀) sty(Ch 9):1.0 ± 0.4%	♂ 59.7 ♀ 61.6	♂ 63.5% alt ♀ >60% alt	lethal
T(8;9)2c[d]	good	ru(Ch 9):1.1 ± 0.6% st(Ch 9):7.2 ± 2.4% (♀)	♂ 58.2 ♀ 51.1	♂ 61.5% alt ♀ random	lethal
T(3;12)	good	hd(Ch 3):4.6 ± 3.2% (♂) 1.4 ± 0.8% (♀) fs(Ch 12):8.9 ± 3.5% (♂) 23.6 ± 3.3% (♀) Pb(Ch 12):43.6 ± 4.6% (♂) 46.2 ± 7.6% (♀)	♂ 60.7 ♀ 61.6	♂ 72.2% alt ♀ >70% alt	lethal
T(7;12)	good	or(Ch 7):3.0 ± 2.1% (♂) 15.7 ± 3.3% (♀) fs(Ch 12):7.5 ± 3.0% (♂) 23.0 ± 4.5% (♀)	♂ 50.6 ♀ 58.3	♂ random ♀ >60% alt	viable but apparently sterile
T(4;8;10)	good	ro(Ch 10):0.5 ± 0.3% y:7.5 ± 2.4% (♂) 8.1 ± 2.0% (♀) cv:complete (♂) 3.0 ± 0.1 (♀) bu:37.7 ± 4.8% (♂) 47.4 ± 2.8% (♀)	♂ 24.0 ♀ 28.0	♂ random --	intercrosses sterile due to embryonic trapping

Table 6. (continued)

Stock	♂ competitiveness	Linkage estimates[a] and, if known, the chromosome carrying the mutant marker	Hatch (%)	Disjunction[b]	Homozygote
T(11;12)	good	fs(Ch 12):9.0 ± 3.1% (♂) 20.4 ± 3.4% (♀) Pb(Ch 12):38.4 ± 4.4% (♂) 46.9 ± 5.6% (♀)	♂ 65.0 ♀ 65.0	♂ 67% alt ♀ >60% alt	intercrosses inconclusive: suggest lethal in males but not in females
T(9;10)7i	good	ro(Ch 10):complete ru(Ch 9):complete	♂ 59.9 ♀ 48.3	♂ 61.2% alt ♀ random	lethal
T(6;8)7h	poor	Bl:0.7 ± 0.7%	♂ 46 ♀ 48	♂ random ♀ random	intercrosses sterile, apparently due to embryonic trapping
T(6;8)2e	nearly equal w.t.	Bl:2.1 ± 1.1%	♂ 45 ♀ 51	♂ random ♀ random	intercrosses sterile

[a] Recombination given separately for the sexes when either the data were from one sex only or there was a sex difference.

[b] Measured from counts of metaphase I cells in males, but estimated from hatch data in females (see text).

[c] Based on data from Fox (1975).

[d] Originally identified as T(9;11) but chromosome 11 was apparently misidentified (Ross and Cochran 1976).

understanding of factors that affect disjunction in translocation heterozygotes. One of us (DGC) discovered that two types of alternate orientation were distinguishable in most of our stocks, in addition to the well known adjacent-1 and adjacent-2 types (Cochran 1977, unpubl.). This adds a new dimension to the chromosome analyses, for it is already evident there are surprising variations within the orientation patterns of different interchanges. Other findings included the location of centromeres in the majority of the chromosomes as the points of earliest diplotene separation (Cochran and Ross 1977, Ross and Cochran 1975, unpubl.). Chromosome identification was facilitated by the recognition of distinctive staining patterns in certain autosomes, undoubtedly reflecting the underlying chromomere patterns. These basic cytogenetic studies were useful in accounting for the differing properties found among the translocation heterozygotes. They are valuable to plans for manipulating stocks to achieve maximum sterility and lethality while simultaneously maintaining a productive breeder system (see Obj. 3).

Crosses between translocation heterozygotes were used to test for a viable homozygote. The mutant marker was expected to show a 3:1 segregation if the homozygote was present, in contrast to a 2:1 if it was lethal. As shown in Table 6 (last col.), positive results were obtained only for T(7;12). However, subsequent attempts to isolate the homozygote were unsuccessful, apparently due to sterility. The other tests either showed a 2:1 segregation or they were completely unproductive. Indeed, all the intercrosses showed high numbers of unhatched egg cases. These led to the first recognition of a phenomena we now believe can be used to develop a unique type of sterile male (Obj. 3). High lethality as in the intercrosses or in double translocations reduces the numbers of viable embryos within the egg case. Sterility from embryonic trapping results when their combined strength is insufficient to force open the keel at the time of hatch (Ross and Cochran 1976). A very precise comparison of lethality vs. sterility from individual oothecae is nearly completed (Kiel and Ross, unpubl.). It is already apparent that 1) increases in lethality between 60 and 80% cause sharp increases in sterility and 2) lethality of 80%, within 10% confidence limits, causes 100% sterility.

The work completed under this objective broadened considerably our understanding of the characteristics of translocation heterozygotes in B. germanica. Six and possibly 8 (2i and 2a), of the stocks were developed into tools of potential value to control measures. As a result of the discovery of embryonic trapping and the knowledge that competitive double translocations can be synthesized (Obj. 3), we have come to the conclusion the most promising use of the translocations is to develop double translocation-bearing males which carry sterilities from 90-100%.

Objective 3 - Laboratory experiments in populations suppression (and the studies of double translocations)

The original plan of work under this objective was tentative. As noted, the selection of the best tools for population suppression was dependent on the results of the analyses of the single translocations (Objective 2). We thought they might be used most effectively in sequential releases of different translocations. It was realized that doubles would

have a more immediate impact on population growth, but we questioned whether the genetic load engendered in the process of synthesis would prohibit the development of a vigorous stock. However, the synthesis and analysis of a double translocation stock showed it was possible to develop fully competitive double males. These studies are summarized in this section, following the information concerned with population studies of single translocations.

A. Population studies with single translocations. Several experiments were made using repeated releases of translocation-carrying males into laboratory populations (Ross 1975, 1976). The experimental and control populations were kept below the carrying capacity of the environment (except for some overcrowding an consequent mortality of small nymphs in mo. 5-6 in the first experiment). Thus, the differences in population size and growth could be used as a measure of the effects of the translocation.

The experiments used two different methods. One consisted of releasing 5th-6th instar T/+ males at about monthly intervals, along with monthly censusing of the population (Ross 1975). T(8;9), originally identified as "T(9;11)", was chosen for the experiment since it showed the best competitiveness of the 3 stocks available at that time. It was demonstrated that the translocation could be successfully incorporated into the population by careful timing of releases. The T(8;9) males showed good competitiveness within the population. They effectively retarded population growth, but nevertheless the population increased considerably in size within the 6 months period of study (Fig. 1). The origin and growth of nymphal groups both within the experimental and control population (*Obj.* 1) were analyzed and insight was thus gained into the dynamics of a laboratory population. Certain groups could not be distinguished in the last months due to overlapping hatch. A better estimate of these was gained using a progeny-group technique (see below).

The second type of experiment used releases into separated progeny groups (Ross 1976). Thus, each nymphal group that appeared within a population jar was separated from its parents, counted, and placed in a fresh jar. A time chart for each group, including dates of hatch and maturation and the instar of the majority of nymphs at monthly intervals, was used to reconstruct the population for each month of growth. Releases were made into each progeny group when the nymphs were in the last instars (5th-6th). Two translocations were tested by the progeny-group technique: T(9;11) carrying a recessive mutant in repulsion (T +/+ st) and T(4;8;10). The latter is a progressive interchange involving 3 chromosomes (Ross and Cochran 1977). One type of double translocation, developed from translocations having one chromosome in common, also involves 3 chromosomes. T(4;8;10) resembles them in respect to lethality, but matings result in a 1:1 ratio of T(4;8;10) to normal (+/+) progeny, whereas offspring of the other three chromosome doubles are all translocation heterozygotes (see next section).

The males for release in the T(8;9) experiment were obtained from a backcross system using the chromosome 9 marker, notched sternite (st). Recombination from crosses of the hybrid females was estimated at 7.2 $\pm$ 2.4%.

Reciprocal test crosses were not made due to sterility of homozygous st females (st/st). It was assumed hybrid males would show similar crossover frequencies since no marked sex differences in crossingover had been found in other studies involving chromosome 9 loci. Thus, it was estimated that 93% of the released males were T +/+ st. Successful matings of males introduced into F_1 progeny groups would then be expected to result in ca. 23% st homozygotes within F_2 nymphal groups.

Releases and mating types in 3 replicates of the T(8;9) experiments are summarized in Table 7. Table 3 lists the progeny groups, of which certain F_2 groups are also shown in the last col. of Table 7. From the latter, it is clear that 2 main factors control the size of the progeny group: the number of normal (+/+) matings and the no. of females in the parental group. The progeny groups in replicate C are much smaller than those in A and B. Sterile egg cases occurred frequently, indicating a lethality beyond that which could be attributed to T(8;9). We cannot explain these results, and can only note that these data are not representative of the effects to be expected from releases of T(8;9), +/st males. Also unexpected were the very low numbers of st phenotypes among the F_2 progeny groups (Table 3). These formed 5-8% of the 3 main sets of F_2 groups in A and B, in contrast to the prediction of ca. 23%. It is possible F_1 males, genotypically t +/+ +, that hatched within the population outcompeted the released males. Another source of phenotypically normal progeny might be matings that included a genotypically normal crossover type (++/++). These two possibilities are the most likely explanations for the low numbers of st progeny.

The populations reconstructed from the progeny groups are shown in Table 8. Replicates A and B were averaged, but data from C are given separately for reasons already noted. Growth in A-B was limited primarily by the translocation. During the 7th month, sterility of F_2 st/st females did not cut hatch by more than 200-300. A reduction of ca. 1000 was estimated on the basis of the expected frequency of st/st (23%). The growth pattern was similar to that of the control, with sharpest increases associated with the first hatch of a new generation (mo. 4 and 7, Tables 4 and 8). As in the first experiment, in which T(8;9) did not carry a deleterious gene, the experimental population was held to about ½ that of the control through the 6th month. It dropped to 1/8 in mo. 7 but, as already noted, this was due largely to the effect of the translocation alone. These experiments showed conclusively that an ordinary single translocation would be incapable of suppressing population growth, at least within a population which was not restricted by environmental factors.

The only single translocation that approached population suppression was the progressive interchange, T(4;8;10). Releases and mating types are summarized in Table 9. Progeny groups from 4 replicates are listed in the last 4 cols. of Table 3. High frequencies of sterile egg cases from embryonic trapping, as well as lethality from unbalanced gametes, contributed to the low numbers within these groups. The effect of the frequency of normal matings on F_2 groups is seen in Table 9, cols. 6 and 7. It is even more pronounced in F_3 groups (Table 10). As shown in Table 9, release ratios of 8-10 T/+:1 +/+ males virtually eliminated +/+ matings. Unfortunately, the value of the higher ratio was not realized until late in the experiments.

Table 7. Summary of releases, their success and effects on progeny groups in T(?;9),+/st experiments with the German cockroach.

Experiment	Group and no.	No. released	Approx. ratio of T/+:+/+ ♂[a]	No. matings (=no. adult ♀)	% +/+ matings	F_2 progeny (group[b] and no.)
A	F_1-1: 78	60	4:1	31	19	F_2(1-1): 512
	F_1-2: 83	85	5:1	41	5	F_2(2-1): 485
	F_1-3: 84	65	4:1	34	21	F_2(3-1): 640
	F_2(1-1): 512	530	5:1	201	2	-----
B	F_1-1: 107	85	4:1	46	9	F_2(1-1): 656
	F_1-2: 112	130	5:1	51	6	F_2(2-1): 581
	F_1-3: 99	105	5:1	38	5	F_2(3-1): 483
	F_2(1-1): 656	738	5:1	236	4	-----
	F_2(3-1):483	375	4:1	201	12	-----
C	F_1-1: 80	98	6:1	28	0	F_2(1-1): 11
	F_1-2: 35	51	6:1	16	0	F_2(2-1): 57
	F_1-3: 59	75	6:1	23	9	F_2(3-1): 22
	F_2(1-1): 118	120	5:1	52	4	-----

[a] Estimated on the assumption that 1/4 of each progeny group were T/+ males.

[b] Groups from 1st egg cases only.

Table 8. - Population[a] growth and structure from experiments using T(8;9),+/st in B. germanica.

Experiment	Age[b]	Month of growth 1	2	3	4	5	6	7
A-B (avg)	Ad	10	10	87	179	235	710	1607
	Lg		77	81	76	485	917	1401
	Md							
	Sm	93	98	92	584	1104	1524	3665
	Total	103	185	260	839	1824	3154	6673
C	Ad	10	10	74	103	146	244	392
	Lg		66	29	49	98	175	295
	Md							
	Sm	80	35	59	118	211	355	648
	Total	90	111	162	270	455	774	1335

[a] Population reconstructed from isolated progeny groups (see text for explanation).

[b] Age is indicated by "Ad" for adult and "Lg" for large (instars 5-6), "Md" for medium (instars 3-4) and "Sm" for small (instars 1-2) nymphs.

Table 9. - Summary of releases, their success and effects on progeny groups in T(4;8;10) experiments with the German cockroach.

Experiment	Group and no.	No. ♂ released	Approx. ratio of T/+:+/+ ♂[a]	No. matings (=no. adult ♀)	% +/+ matings	F_2 progeny (group[b] and no.
A	F_1-1: 34	43	6:1	14	16	F_2(1-1): 101
	F_1-2: 15	20	5:1	12	20	F_2(2-1): 117
	F_1-3: 27	40	6:1	16	6	F_2(3-1): 71
	F_2(1-1): 101	76	4:1	54	6	----
	F_2(2-1): 117	112	5:1	51	6	----
	F_2(2-2): 123	120	5:1	54	6	----
	F_2(3-1): 71	80	5:1	36	8	----
	F_2(3-3): 87	109	6:1	41	2	----
B	F_1-1: 11	12	5:1	8	0	F_2(1-1): 42
	F_1-2: 45	48	5:1	10	20	F_2(2-1): 119
	F_1-3: 14	18	6:1	9	11	F_2(3-1): 52
	F_2(1-1): 42	50	6:1	20	5	----
	F_2(1-2): 24	42	8:1	11	0	----
	F_2(1-3): 23	53	9:1	9	0	----
	F_2(2-3): 37	65	8:1	16	0	----
	F_2(3-1): 52	98	8:1	21	5	----
	F_2(3-2): 54	110	9:1	21	0	----
C	F_1-1: 43	54	6:1	17	12	F_2(1-1): 152
	F_1-2: 62	78	6:1	24	8	F_2(2-1): 114
	F_1-3: 41	51	6:1	18	6	F_2(3-1): 73

Table 9. - (continued)

Experiment	Group and no.	No. ♂ released	Approx. ratio of T/+:+/+ ♂[a]	No. matings (=no. adult ♀)	% +/+ matings	F_2 progeny (group[b] and no.)
C	F_2(1-1): 152	185	6:1	52	2	----
	F_2(1-3): 42	95	10:1	20	5	----
	F_2(2-1): 114	142	6:1	52	10	----
	F_2(2-2): 96	200	10:1	24	0	----
	F_2(3-1): 73	164	10:1	33	0	----
D	F_1(1-1): 47	50	5:1	17	6	F_2(1-1): 127
	F_1(1-2): 31	55	8:1	14	0	F_2(2-1): 58
	F_1(1-3): 14	32	10:1	8	0	F_2(3-1): 28
	F_2(1-1): 127	212	8:1	42	2	----
	F_2(1-2): 68	120	8:1	24	8	----
	F_2(2-1): 58	101	8:1	24	4	----

[a] Estimated on the assumption that 1/4 of each progeny group were T/+ males.

[b] Groups from 1st egg cases only.

Table 10. - Mating history of F_3 groups from T(4;8;10) experiments with B. germanica.

No. +/+ F_1 matings	No. +/+ F_2 matings	No. progeny/F_3 group[a] (control group = ca. 32000)
0	0	0,0
		20
		21, 10
	1	65
		68
1	0	21
		51
	1	82
		147
		135, 96
		69, 109, 78
	2	158
	3	269, 274, 212
2	0	60[b]
		107
	1	154[b]
		386
	2	--
	3	595, 641, 336
		495, 396
		487, 274
	4	--
	5	457
		529

[a] Counts from 2nd and 3rd progeny groups, where available, follow the 1st progeny group.

[b] From small F_2 parental groups (3rd egg case progeny groups).

Table 11. Population[a] growth and structure from experiments using T(4;8;10) in B. germanica.

Experiment	Age[b]	Month of growth						
		1	2	3	4	5	6	7
A-C (avg)	Ad	10	10	34	64	84	181	326
	Lg		24	33				
	Md				23	97	160	186
	Sm	29	41	27	117	195	224	579
	Total	39	75	94	204	376	564	1091
D	Ad	10	10	44	70	80	185	380
	Lg		39		12			
	Md			26		105	105	105
	Sm	47	31	14	127	126	126	195
	Total	57	80	84	209	311	416	680

[a]Population reconstructed from separated progeny groups (see text for explanation).

[b]Age is indicated by "Ad" for adult and "Lg" for large (instars 5-6) "Md" for medium (instars 3-4) and "Sm" for small (instars 1-2) nymphs.

Ratios were increased in some late-hatching progeny groups, of which the effects on the first F_3 groups were seen only in replicate D (Table 11, mo. 7).

The T(4;8;10) populations as reconstructed for 7 mo. of growth are shown in Table 11. One was based on means from replicates A-C. The other, Pop. D, is shown separately due to differences in release ratios. In mo. 7, the total numbers in A-C and D were 2.1% and 1.3% of those estimated for the control. Higher releases at the initiation of the experiment would have improved upon these results. The advantage over the 2-chromosome translocation is evident in the comparison of both the corresponding progeny groups (Table 3) and the reconstructed populations (Tables 2 and 11). These experiments leave little doubt that the ordinary reciprocal translocation would be incapable of suppressing growth of field populations unless aided by special environmental factors. In contrast, T(4;8;10) might be useful for keeping growth at very low levels for periods of 6-7 months.

B. Synthesis and study of double translocation heterozygotes. The advantage of most double translocations over T(4;8;10) stems from differences in progeny genotypes, as follows:

Mating	
T(4;8;10) X +/+	1 T/+:1 +/+
4-chromosome dbl. X +/+	1 T^1/+:1 T^2/+:1 T^1, T^2; 1 +/+
3-chromosome dbl. X +/+	1 T^1/+:1 T^2/+

Since matings (releases) of T(4;8;10) leave 50% of the progeny free of a translocation, in contrast to 25% or none, use of the doubles will obviously have a greater impact on population growth. The greatest potential would seem to lie with the 3-chromosome double, since all progeny would be heterozygous for one or the other parental translocation. However, synthesis of a combination characterized by lethalities of 75% or more might cause such high sterilities that it would be preferred to a 3-chromosome double of lesser lethality.

The first hatch data from double translocations were from two 4-chromosome doubles that included T(2;11)Cu (Ross and Cochran 1976). This type combines two independent translocations and hence it involves 4 chromosomes. Matings of the double males to wild-type females reduced the progeny averages/1st ootheca from a normal of 40.0 to 5.0 and 0.2 nymphs, respectively. This was due to the combined effects of lethality and sterility from embryonic trapping. It appeared that double stocks developed from translocations which, unlike T(2;11)Cu, had good male competitiveness, might be capable of population suppression.

T(8;9) and T(3;12) were two of the first single stocks for which identification by closely linked visible markers was established, and which also exhibited good male competitiveness (Table 6). Therefore, these were used as the first double for which a detailed analysis was made. The stock was developed using a double mutant stock of ru on chromosome 9 in T(8;9) and hd on chromosome 3 in T(3;12). Crosses between appropriately marked T(8;9) males and T(3;12) females proved to be the best source of doubles, as they averaged 5.6 nymphs/ootheca (1st) in comparison to 3.8 in the reciprocal cross. Phenotypically normal males from the intercross system were checked cytologically and, as expected, two ring or cross

configurations occurred in pachytenediplotene cells, verifying the presence of both translocations. The results of 50 1:1 male competitiveness tests showed the double males were equally competitive with wild-type laboratory males. Their matings averaged 2.5 ± 0.7 nymphs/1st ootheca, with 70% sterility among 1st and 2nd egg cases. This increased in later ootheca.

The sterility associated with matings of the double males was higher than expected since both T(3;12) and T(8;9) males are normally characterized by a favoring of the viable chromosome combinations, i.e., directed alternate disjunction. Examination of cells from double males showed that disjunction in T(3;12) had changed from directed to random. This increased the lethality and explains the high levels of sterility. This type of situation can be manipulated to obtain satisfactory intercross systems for producing doubles which themselves carry extremely high sterilities.

The 2nd double to be synthesized was T(7;12), T(8;9). The characteristics, including good competetiveness, were closely similar to the T(3;12), T(8;9) double. A mean sterility among 1st oothecae of 70% was again associated with the occurrence of random disjunction in one translocation and directed in the other. Surprisingly, it was T(8;9), rather than T(7;12), that changed to random segregation. The identification system used ru on chromosome 9 and, for T(7;12), the marker was orange-body (or) on chromosome 7. Intercrosses for the production of the double used T(7;12) males. T(7;12) females were unsatisfactory due to a higher crossover rate for or. This system was somewhat less productive than that for T(3;12), T(8;9) doubles, but, on the other hand, or is more readily identified than the hd marker used in T(3;12), T(8;9) synthesis. Because of this, it would be difficult to choose between these two doubles if they represented the most promising of available mechanisms. However, higher sterilities are associated with several other double translocations as described below.

A third 4-chromosome double was developed late in the Contract period. It combined T(7;12) and T(9;10)7i. The identifying markers were or on chromosome 7 and rose-eye on chromosome 10. Within the available time of study, it was only possible to acquire data on hatch and sterility. Matings of the double males resulted in 90% sterility among 1st oothecae, with progeny averages of only 0.3. Since the lethality is clearly higher than in either the T(3;12), T(8;9) or the T(7;12), T(8;9) double, we suspect both translocations may have changed to random disjunction within the double males. The T(7;12), T(9;10)7i double provides an alternative to the three double stocks we are currently considering for field studies.

The requisite stocks to develop a 3-chromosome double type, T(3;7;12), are available. This system combines translocations with one chromosome in common, e.i. T(3;12) with T(7;12). The identification system requires a mutant marker which is not on the common chromosome. T(3;7;12) was synthesized using hd on chromosome 3 and or on no. 7. Fifty 1:1 male competitiveness tests gave the following results: 23 +/+ matings; 27 T,T matings. The average sterility from embryonic trapping was 89% and the mean progeny/1st ootheca 0.3, as compared to 70% and 2.4-2.5 progeny in the two better-studied 4-chromosome doubles. A single pair could

replace itself 2-3 times in the latter (progeny from all oothecae); in T(3;7;12), average replacement was less than 1 cockroach. Another advantage over the 4-chromosome type is that, due to differences in chromosome segregation at meiosis, all progeny are heterozygous for one or the other parental translocation. This was confirmed by chromosome studies. All male progeny were heterozygous for either T(3;12) or T(7;12). T(3;7;12) is a mechanism worthy of field testing, although it would be desireable to have at least some prior data on a laboratory population study.

The recent discovery that black-body (Bl) is located on chromosome 8 in T(8;9) opens new possibilities for the use of this translocation. With Bl on 8 and ro on 10, T(8;9) could be combined with T(9;10)7i to form a 2nd 3-chromosome double, T(8;9;10)7i. It would be worthwhile to determine whether such a stock would have any advantage over T(3;7;12). The same markers could be used to develop a stock of T(8;9) with T(4;8;10). In crosses to check on the common involvement of chromosome 8, productive matings were obtained between T(8;9) males and T(4;8;10) females. Apparently this was possible due to strong directed disjunction in the females plus directed disjunction in T(8;9) males. As expected, lethality in the reciprocal crosses resulted in complete sterility from embryonic trapping (ca. 84% lethality). Double males from the productive intercross were examined cytologically. The translocations showed separate configurations. Apparently chromosome 8 was mis-identified in T(4;8;10), but the designation will not be changed until we are certain of the correct identification. More importantly, both translocations showed their normal disjunction characteristics, i.e., random in T(4;8;10) and directed in T(8;9) males. The overall lethality was estimated at 80%. This is the level at which we expect near to complete sterility from embryonic trapping. If competitive, these sterile males will be the mechanism of choice for field releases.

C. A population study using the T(3;12), T(8;9) double. The progeny-group method was used to study the effects on releasing T(3;12), T(8;9) males on population growth. As in the other experiments, the populations were started with 5 +/+ females mated to the translocation-carrying males. Releases into subsequent nymphal groups were between 1.5 and 2X the number within the group. Five replicates were set up. The average size of each group up to and including the last hatch of an F_3 group is shown in Table 12. This table is organized so that the origin of each group can be traced back to one of the 4 F_1 progeny groups, i.e., those hatching from the 1st 4 egg cases of the parental group. There was no hatch from 5th egg cases. The totals of all nymphs produced in each generation are as follows:

F_1: 34.4
F_2: 67.2
F_3: 43.9

Thus, population decline was initiated in the 3rd generation.

The population as reconstructed for the first 11 mo. of growth is shown in Table 13. The numbers of small nymphs reflect the pattern of growth somewhat ahead of the total numbers as they are the future breeding groups. However, both show a strong retardation of growth, followed by leveling off and eventual decline. The latter corresponds to the reduction in F_3 groups noted above. The experiment confirms our predictions that a

Table 12. Progeny groups averaged from 5 replicates of T(3;12), T(8;9) population experiments.

F_1 groups[a]	F_2 groups	F_3 groups	
F_1-1: 7.0	F_2(1-1): 12.4	F_3(1-1-1): 7.8	F_3(1-1-3): 2.8
		F_3(1-1-2): 6.2	
	F_2(1-2): 10.4	F_3(1-2-1): 0.8	F_3(1-2-3): 0.8
		F_3(1-2-2): 2.0	
	F_2(1-3): 4.4	complete sterility	
	F_2(1-4): 2.4	complete sterility	
F_1-2: 12.8	F_2(2-1): 9.4	F_3(2-1-1): 5.8	F_3(2-1-3): 2.6
		F_3(2-1-2): 5.6	
	F_2(2-2): 11.0	F_3(2-2-1): 2.2	F_3(2-2-3): 1.2
		F_3(2-2-2): 3.6	F_3(2-2-4): 1.0
	F_2(2-3): 6.0	F_3(2-3-1): 1.4	
	F_2(2-4): 4.6	complete sterility	
F_1-3: 6.8	F_2(3-1): 1.8	complete sterility	
	F_2(3-2): 1.6	complete sterility	
	F_2(3-3): 3.2	complete sterility	
F_1-4: 0.6	complete sterility		

[a]Dotted lines indicate origins of progeny groups. For example, the first F_1 group (hatch from 1st egg cases) produced F_2 progeny from 4 successive egg cases and these, in turn, gave rise to 6 F_3 groups.

Table 13

(Studies using a 4-chromosome type of double translocation)

Average growth of a laboratory population following sequential releases of a double translocation heterozygote, T(3;12), T(8;9), from 5 replicates.

Age	Month of Growth 1	2	3	4	5	6	7	8	9	10	11
Ad	10	10	15.8	26.4	22.0	32.8	49.4	57.8	62.0	70.5	70.1
Md-Lg		5.8	10.6	5.6	10.8	16.6	14.3	14.8	14.1	8.5	4.5
Sm	7.0	12.8	6.8	13.0	20.0	17.2	17.8	17.0	12.6	6.4	5.0
Total	17.0	28.6	33.2	45.0	52.8	66.6	81.5	89.6	88.7	91.1	79.6

Total No. of 1st generation nymphs (F_1): 34.4

2nd generation nymphs (F_2): 67.2

3rd generation nymphs (F_3): 43.9

4-chromosome double was potentially capable of suppressing population growth. The higher sterilities and lethalities associated with certain other doubles, such as T(3;7;12), indicate suppression could be achieved more rapidly with one of these mechanisms.

Objective 4 - Field Trials. It was not possible to undertake these within the given time frame.

Concluding Remarks

The past three years of research have brought us close to the point at which we feel field tests should be conducted. The mechanism would be one of the double translocations which show 90-100% sterility and good competitiveness. The most effective use of translocations in B. germanica almost certainly lies in the development of such stocks. The sterility from embryonic trapping approaches and, indeed, may equal that produced by standard methods such as radiation or chemosterilants, while avoiding the debilitating effects of such treatments. A sufficient number of single translocations were analyzed to give considerable flexibility in the synthesis of double translocations. It is noteworthy that the males of the three best-studied doubles showed good competitiveness, even though heterozygous for two translocations and two mutant markers. Apparently, a rather large genetic load can be carried in the heterozygous state without detectable loss of fitness.

Basic cytogenetic research constituted an important component of the studies completed under the Contract. Mutant markers had to be found which were sufficiently close to chromosome breakpoints to provide reliable means of identification. Chromosome disjunction data aided in the selection of suitable backcross and/or intercross systems for both the single and double translocations, as well as in understanding hatch and other characteristics. It was discovered that disjunction properties of certain translocations altered with the genetic backgroud. These changes provided the explanation for unexpectedly high sterilities in several double males. They revealed possibilities for manipulating disjunction properties to enhance the productivity of source systems while achieving maximum sterility in males for release. There are also broader implications, both in respect to the use of translocations for control in other insects and factors that affect chromosome segregation in translocation heterozygotes.

It seems the main problem in using genetic control techniques lies in objections to releasing cockroaches. Other parameters that might mediate against successful application, such as behavioral or genetic differences between free and laboratory-bred insects or the inability to locate suitable release sites, do not appear to be particularly significant for B. germanica. The studies of field collections were limited, yet they suggested the releases might be relatively small. If a population was reduced by a non-residual insecticide, followed by releases of fewer insects than in the original population, some of the objections might be overcome. Possibly, too, people are becoming more aware of both the adverse environmental effects and personal health hazards associated with pesticides.

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Cochran, D. G. Disjunction types and their frequencies in two heterozygous reciprocal translocations of Blattella germanica (L.). Chromosoma 59:129-135. 1976.

Cochran D. G. and M. H. Ross. Cytology and genetics of a stable ring-of-six translocation in the German cockroach. J. Hered. (in press).

Ross, M. H. Use of a translocation heterozygote to retard a laboratory population of the German cockroach. Environ. Entomol. 4:37-45. 1975.

Ross, M. H. Genetic variability in the German cockroach. X. Genetics of pale purple, pearl and stumpy. J. Hered. 66:155-159. 1975.

Ross, M. H. Laboratory population studies of the German cockroach using a two-chromosome and a three-chromosome reciprocal translocation. Ann. Entomol. Soc. Amer. 69:1073-1081. 1976.

Ross, M. H. and D. G. Cochran. Sterility and lethality in crosses involving two translocation heterozygotes of the German cockroach, Blattella germanica (L.). Experientia 32:445-447. 1976.

Ross, M. H. and C. H. Wright. Characteristics of field-collected populations of the German cockroach. Proc. Wash. Ent. Soc. (in press).

In addition, three manuscripts are currently under preparation.